STATE OF CALIFORNIA

DEPARTMENT OF TRANSPORTATION

CONTROLLING THE SPREAD OF *XYLELLA FASTIDIOSA*, THE CAUSAL AGENT OF OLEANDER LEAF SCORCH, BY DISRUPTING VECTOR ACQUISITION AND TRANSMISSION

REPORT NO. FHWA/CA/OR-02-01

CALTRANS STUDY NUMBER
(1 DECEMBER, 2001 THROUGH 30 JUNE, 2002)

FINAL REPORT

JULY, 2002

1. REPORT NO.	2. GOVERNMENT ACCESSION NO.	3. RECIPIENT'S CATALOG NO.
FHWA/CA/OR-02-01		

4. TITLE AND SUBTITLE

CONTROLLING THE SPREAD OF *XYLELLA FASTIDIOSA*, THE CAUSAL AGENT OF OLEANDER LEAF SCORCH, BY DISRUPTING VECTOR ACQUISITION AND TRANSMISSION

5. REPORT DATE

July 2002

6. PERFORMING ORGANIZATION CODE

7. AUTHOR(S)

Blua, M. J., Redak, R. A., and Costa H.S.

8. PERFORMING ORGANIZATION REPORT NO.

9. PERFORMING ORGANIZATION NAME AND ADDRESS

California Department of Transportation
Research & Innovation, MS#83
P.O. Box 942873
Sacramento, CA. 94273-0001

10. WORK UNIT NO.

11. CONTRACT OR GRANT NO.

N/A

12. SPONSORING AGENCY NAME AND ADDRESS

California Department of Transportation
Sacramento CA. 95819

13. TYPE OF REPORT & PERIOD COVERED

Final Report

14. SPONSORING AGENCY CODE

15. SUPPLEMENTARY NOTES

This project was performed in cooperation with the US Department of Transportation, Federal Highway Administration.

16. ABSTRACT

Oleander Leaf Scorch (OLS) is a devastating disease of oleander induced by the bacterium *Xylella fastidiosa*. The Glassy-winged Sharpshooter (GWSS, *Homalodisca coagulata*) spreads the OLS bacterium during feeding by first acquiring it from infected plants and then inoculating non-infected oleanders. Currently, OLS is devastating oleanders from San Diego to Ventura counties along the cost, and inland to San Bernardino and Riverside counties.

Our research involves developing a management strategy for OLS that uses a systemic insecticide to interfere with GWSS acquisition of *X. fastidiosa* from infected oleanders and their ability to inoculate non-infected oleanders.

Our study shows that applications of Merit (imidacloprid) induce GWSS mortality on OLS-diseased and healthy oleanders, and have a residual activity in excess of 15 weeks. Our experiments were inconclusive regarding the impact of Merit on the acquisition of the OLS bacterium from treated-infected oleanders, and its inoculation by infectious GWSS into treated-healthy oleanders.

17. KEY WORDS	18. DISTRIBUTION STATEMENT
Homalodisca coagulata, *Homalodisca lacerta*, Sharpshooter, *Xylella fastidiosa*, Oleander, Oleander leaf Scorch, insecticides	No Restrictions. This document is available through the National Technical Information Service, Springfield, VA 22161

19. SECURITY CLASSIF. (OF THIS REPORT)	20. SECURITY CLASSIF. (OF THIS PAGE)	21. NO. OF PAGES	22. PRICE
Unclassified	Unclassified	14	

Table of Contents

List of Figures

DISCLOSURE

CALTRANS entered into a research agreement (Master agreement No. 65Y350) with the University of California, Riverside to contribute to their Entomology Department's ongoing research project to find management strategies for sharpshooter vectors of Oleander Leaf Scorch. CALTRANS contributed $47,686 towards this effort. The enabled the Entomology Department to hire a part-time Assistant Research Entomologist and a part-time Student Assistant to work on this project, and purchase supplies necessary for this project.

DISCLAIMER

Acknowledgments

We would like to thank Bayer Corporation (Kansas City, MO) for providing Merit.

Introduction

This report documents experiments we conducted to determine the impact of a soil-applied systemic insecticide, Merit 75 WP (Imidacloprid, Bayer Corp. Kansas City, MO), on the transmission of a plant-pathogenic bacterium, *Xylella fastidiosa*, to oleander by the glassy-winged sharpshooter (GWSS).

Oleander leaf scorch (OLS) is a disease of oleander induced by a strain of *X. fastidiosa*. The disease causes unsightly "scorch" symptoms that are consistent with a chronic and progressive water stress. This disease kills plants within about two years after the first appearance of symptoms. OLS was first detected in Orange County and the Palm Springs area in the mid 1990's and has spread throughout southern California. Currently, OLS is destroying oleander in San Diego, Orange, Riverside, San Bernardino, Los Angeles, and Ventura counties. Oleander is a mainstay of CALTRANS right-of-way planting, and cannot be easily replaced. A 1997 estimated economic impact of the loss of oleanders on state highways alone is $75 million, with plant replacements adding another $50 million (Henry, et al. 1997).

Other strains of *X. fastidiosa* cause serious diseases in grape, peach, citrus, almond, numerous ornamental tree species, and other crops in many parts of North and South America (Hopkins 1989). The strain that induces OLS is distinctive and appears to be new to California on the basis of DNA analyses (Hendson et al. 2001).

Leafhoppers known as the glassy-winged sharpshooter (GWSS), *Homalodisca coagulata*, and the smoke tree sharpshooter, *Homalodisca lacerta*, are the primary vectors of the OLS bacterium in California. The former insect is native to the southeastern United States where it is a principle vector of phony peach disease and Pierce's disease of grapevines, which are caused by other strains of *X. fastidiosa* (Hopkins 1989). *H. coagulata* was first detected in California in Orange and Ventura counties in 1989 (Sorensen & Gill 1996) and is now as far north as Sacramento County. It is well established and thrives in the Los Angeles basin. A closely related species, *H. lacerta* is native to the drier areas of California. Its spatial distribution and host range overlaps to a large degree with *H. coagulata.* Eradication of these vectors is not feasible. Unlike other sharpshooters, both of these insects can reproduce on oleander. This could allow OLS to disseminate throughout California along highway plantings of oleander.

The *X. fastidiosa*-sharpshooter-oleander system has several factors that make disease control difficult. Symptoms of the disease may not appear for several months, and there is no known way to cure infected plants, which eventually die from infection. Infected plants, even without symptoms, are point sources from where the pathogen is spread by sharpshooter. The vectors are extremely mobile, and have a large host range that include many common ornamental and crop plants. These factors, and the nature of studying a disease that is the result of a three-way interaction among divergent organisms, inhibits experimentation and quick solutions.

If OLS continues to spread an important part of landscape horticulture in the southwest will be lost. Landscape professionals agree that oleander will be difficult to replace. Oleander is frequently overlooked and under-appreciated because it is common, drought tolerant, and requires little care. It can take different forms, from low shrubs to small trees, and blooms with different colors, including pink, salmon, red, white, and yellow. Oleander is used primarily as an ornamental, but also plays an important role as windbreaks, borders, and right-of-way plantings. The California Department of Transportation (CALTRANS) maintains oleander in over 2,100 miles of freeway median, and oleander is found in 20% of all home gardens in California. Oleander is a mainstay in shopping centers, parks, and golf courses. Besides being important in California, oleander is used similarly in other states of the southwestern United States, including Arizona, New Mexico, Nevada, and Texas.

From November 1996 through October 1999, we trapped sharpshooters semimonthly on oleander and citrus in three locations that have OLS, but represent different ecosystems and vector species complements (Fig. 1 and 2). In Irvine (Orange Co.) *H. coagulata* is the predominant sharpshooter, and it is more prevalent on citrus than oleander. In Riverside, *H. coagulata* and *H. lacerta* are both found, and both are more prevalent on citrus than oleander. In Palm Desert, only *H. lacerta* is found, and it is more prevalent on oleander than citrus. At all three sites, sharpshooter populations begin to rise in late May to early June, peak in July, and declined in August-September. Thus, efforts to protect plants by managing sharpshooter vectors may be concentrated to less than one-half of the year.

There are no therapies available to treat infected plants, and none will likely be developed in the foreseeable future. In general, the most effective control strategies for diseases caused by insect-vectored pathogens involve altering aspects of the host plant to render it tolerant or a non-host for the pathogen. Usually this takes the form of selecting pathogen-tolerant or resistant cultivars, or changing the genetic make-up of the host to make it resistant through breeding programs or genetic engineering. However, other management tactics designed to slow the spread of *X. fastidiosa* may be used immediately to reduce the economic impact of the disease while long-term solutions are sought. Such tactics include using insecticides to control insect vectors. Insecticides may work to reduce disease spread by killing or repelling vectors before they can acquire the pathogen from an infected plant, or transmit the pathogen to a healthy plant. Depending on the specific details of the acquisition and transmission process, and action of the insecticide in question, one or both of these factors may take place.

Presently, details are not known regarding the time required for sharpshooters to acquire *X. fastidiosa* from infected oleander and transmit it to healthy plants. However, for similar systems, several hours of acquisition and transmission access are required for efficient disease spread (Purcell & Finlay 1979). Our laboratory studies under conditions designed to optimize pathogen transmission indicate that *H. coagulata* is an efficient vector for oleander. Rates of transmission using three or one vector per plant were 97% and 75%, respectively (Costa et al 2000).

In our greenhouse studies with oleander, we found that soil-applied systemic insecticides of the chemical class known as chloronicotinyls have a strong impact on the feeding behavior of *H. coagulata*. In a greenhouse experiment, pathogen-carrying sharpshooters released on treated oleanders died in an average of 9 minutes on plants treated with imidacloprid (Bayer Inc), after first feeding contact, while none of the individuals on non-treated oleanders died within 24 h (Fig 3). In addition, the mean number of minutes spent feeding by sharpshooters on treated plants was 3 minutes, while on non-treated plants sharpshooters fed for almost the entire observation period of 240 minutes. Ultimately, none of the treated plants became infected, while 75% of the control plants were positive for infection by the OLS bacterium (Fig. 3).

It is likely that these insecticides affect sharpshooters before they could acquire or transmit the pathogen. Besides affecting sharpshooters immediately, these insecticides have a long residual activity in treated plants. In earlier studies they induced high levels of mortality 3 weeks after application (Fig. 4) (Perring et al. 1997). Long-lasting efficacy will allow long intervals between subsequent treatments, thus mitigating environmental effects and the cost of insecticide application.

There is precedence for insecticides impacting the spread of a disease caused by *X. fastidiosa* in citrus (Adlerz et al. 1989). Currently, chloronicotinyls are being used to control outbreaks of an *X. fastidiosa*-induced disease of citrus in Brazil (A.H. Purcell, personal communication) and grapes in California. In addition, the relative safety of these products is revealed by the fact that they are used frequently in vegetable crops in California, even in areas of dense populations in Orange and San Diego counties. One formulation of one of these insecticides, Admire (Bayre Inc), is registered in California for application through irrigation systems in agricultural products. For CalTrans, further reduction in the cost of insecticide application would be gained if the formulation of imidacloprid for ornamentals was registered for application through the sophisticated irrigation systems already in place in some districts.

Objectives and Scope. The goal of our research is to determine if systemic insecticides can be part of an integrated pest management system to reduce the spread of *X. fastidiosa*, the causal agent of oleander leaf scorch, by sharpshooter vectors. Specific objectives are as follows.

(1) To ascertain the degree to which a systemic insecticide affects the acquisition of *X. fastidiosa* by sharpshooters from diseased oleanders through time after plants are treated.

(2) To characterize the relationship between time that sharpshooters are allowed access to diseased oleanders and the probability that they acquired *X. fastidiosa.*

(3) To ascertain the degree to which a systemic insecticide affects transmission of *X. fastidiosa* to oleanders by pathogen-carrying sharpshooters through time after plants are treated.

Body of Report / Technical Discussion

Materials and Methods

General methods. Our experiments were conducted at the University of California, Riverside, CA. We used field-grown oleanders started from cuttings. A woven mesh material (ca. 3mm mesh size) supported by conduit frames covered experimental oleanders to keep GWSS flying in the area from spreading disease and thus compromising our experiments. GWSS used in experiments were adults collected from citrus groves at the University of California, Riverside, CA.

The systemic insecticide we examined was Merit 75WP, a soil-applied formulation of imidacloprid registered for use on ornamental shrubs. Imidacloprid, in the class of insecticides known as chloronicotinyl, has a low mammalian toxicity and no known activity against phytophagous mites or nematodes, and as a soil-applied formulation can be used with biocontrol (Anon 1994, Elbert and Overbeck 1990).

Bacterial acquisition experiment. We used oleanders (cv Hearty Red) that were mechanically inoculated two years before the experiment with an OLS-inducing isolate of *X. fastidiosa* (American Type Culture Collection 700598). All of these oleanders showed moderate to severe disease symptoms by the time the experiment was initiated. This experiment had two treatments: (1) untreated OLS-diseased oleanders and; and (2) OLS-diseased oleanders that were treated with Merit 75WP. These oleanders were treated on the on the basis of trunk diameter according to the manufacturer's directions. One liter of aqueous solution containing 5.67 gm of Merit 75WP was applied to the soil at the base of the plants while they were undergoing routine irrigation.

Oleanders were treated on 17 July 2001, and each treatment had 8 replicates. Every other week between 26 July 2001 and 15 November 2001 10 field-collected GWSS adults were placed in sleeve cages on 3 separate branches of each experimental oleander and allowed to feed. Sleeve cages consist of 45cm long x 25 cm diameter organdy bags that were used to enclose insects on oleander branches. One cage was removed after 2, 4, and 6 hours, respectively, from each plant. For each cage the number of dead GWSS was noted. Statistical analysis consisted of t-tests to determine significant differences in the numbers of dead GWSS between treatments (SAS Institute, 1999). Data were square-root transformed (Steel and Torrie, 1980). Linear regression was used to examine the number of dead GWSS as a function of days after treatment with Merit (Wesolowsk 1976). Live GWSS were placed in a vial and stored frozen for future assays to determine if they had acquired *X. fastidiosa*.

Bacterial inoculation experiment. This experiment included two factors. The first factor (2 levels) was treatment with Merit 75WP as above on 4 October 2001, or untreated controls. The second factor (3 levels) was time interval between treatment and exposure to infectious GWSS, either 2, 8, or 12 weeks. Experimental oleanders ("Sister Agnes", a cultivar that is very sensitive to OLS) were prepared by rooting cuttings from non-infected plants 8 months before the experiment was initiated. The experiment included 10 replicates for each treatment/interval combination. GWSS adults that were allowed to acquire *X. fastidiosa* from untreated control oleanders that were used in the bacterial acquisition experiment (above). These GWSS were placed in sleeve cages on symptomatic branches and allowed to feed for 4 days to maximize acquisition. Three such GWSS were placed on treated and control oleanders at the designated post-treatment time interval for 48 hours. Experimental oleanders were examined on 30 May 2002 for symptoms of OLS. At the same time, two leaves were collected from the second to third whorl from the terminal on the branch on which infectious GWSS were placed. These leaves were sent to AgriAnalysis (Davis, CA) for determination of infection by *X. fastidiosas* using polymerase chain reaction.

Results

Bacterial acquisition experiment. One week after treatment with Merit, a substantial impact on GWSS mortality was observed in GWSS exposed to treated plants for 4 and 6 hours, but not 2 hours (Fig 5). Greater mortality was observed on treated vs. control oleanders for each date/exposure time combination with the exception of the 2 hr exposure time 1 week after treatment application, when no mortality was observed. Significant differences in the mortality of GWSS between treated and control oleanders were indicated on 5 of 8 dates for 2 hours of exposure, 8 out of 8 dates for 4 hours of exposure, and 7 out of 8 dates for 6 hours of exposure (Fig. 5). No linear tendency (ie slope = 0) was observed in the GWSS mortality induced on treated plants through time after treatment. This indicates that the impact of imidacloprid in oleander on GWSS mortality likely would last beyond the 15 week experimental period. Least squares mean mortality (± standard error) across dates after 2, 4, and 6 hours of exposure was 1.5 (+0.026, 10.022), 2.8 (+0.032, -0.029), and 5.4 (+0.034, -0.034), respectively, for control oleanders. On Merit-treated oleanders least squares mean mortality (± standard error) across dates after 2, 4, and 6 hours of exposure was 4.9 (+0.034, 10.034), 7.3 (+0.029, -0.031), and 9.4 (+0.015, -0.019), respectively (Fig 5). The largest difference in GWSS mortality between control and Merit-treated oleanders was at 4 hours after exposure. Based on least squares mean across date the difference was 45%.

This experiment demonstrated that even symptomatic oleanders take up imidacloprid, the active ingredient in Merit, which induces GWSS mortality. This observation is important for managing GWSS that would otherwise have an opportunity to acquire *X. fastidiosa*. High control mortality reveals that oleander, at least when infected, is not a good feeding host for the GWSS. This is particularly apparent after 6 hours of exposure, and is consistent with our observation that the GWSS is not found in association with oleander relative to others hosts (Blua et al, 2001). The high variability in GWSS mortality 6 hours after exposure is likely due to variable temperatures from July 26 to November 1, 2001, or changes in the age of the insects used. Our previous experience with GWSS indicates that older adults are more sensitive to manipulation than younger ones.

Although GWSS that were alive from this experiment were collected and stored frozen, we declined to assay them for *X. fastidiosa*. Our recent experience with assays for *X. fastidiosa* in GWSS indicates that it is less reliable than we originally anticipated. In addition, the process is destructive to the sample. In cooperation with Dr. D. Cook (U.C. Davis,) we have recently been granted research support from the University of California to better develop methods to examine *X. fastidiosa* in the GWSS. Once a solid assay is developed we will examine the GWSS collected from the bacterial acquisition experiment.

Bacterial inoculation experiment. On the three dates that infectious GWSS were placed experimental oleanders for inoculation studies, an overwhelmingly high percentage of individuals died on Merit-treated oleanders (100%) relative to untreated controls (7.5%) after 24 hours. At the most recent sampling period (6 months after inoculation) none of the inoculated oleanders were showing symptoms of infection by *X. fastidiosa*. In addition, none tested positive for *X. fastidiosa.* We are continuing to maintain these oleanders to check for the development of OLS within the next year.

Evaluation

Unfortunately, we currently cannot answer the most important questions this research posed. First, we do not know the degree to which imidacloprid in infected oleanders blocked the acquisition of *X. fastidiosa* by the GWSS. However, because we have stored GWSS from our experiment to address that question, we can determine if they had acquired *X. fastidiosa* once we develop and test a reliable method. Second, because test plants are not symptomatic, we were not able to determine the degree to which inoculation of the bacterium into oleander by the GWSS was blocked by imidacloprid. We will continue to monitor oleanders used in that experiment. Further examination at a later date may reveal disease symptoms that are not apparent now.

Imidacloprid applied at label rates to infected oleander induced a high level of mortality in GWSS for over 3 months. At the end of the experiment, substantial mortality was induced in treated oleanders. To our surprise there was no decrease in mortality on treated oleanders through the experimental period. This means that the impact of Merit to GWSS on infected oleanders would last longer than we initially projected. We would not expect applications of Merit to oleanders that were not infected to provide any less control.

Merit affected the GWSS rapidly. On the basis of least squared means, after 2 hours of exposure, most of the total mortality had occurred that was induced after 6 hours. A rapid impact on the GWSS is important to disrupting acquisition of *X. fastidiosa* from infected oleander and its inoculation into plants that are not infected. The processes of acquisition of *X. fastidiosa* by other sharpshooters and inoculation of the bacterium into plants requires hours (Purcell Finlay 1979). The effect of Merit may be underestimated by considering only mortality. We do not need to kill insects to keep them from acquiring the bacterium or inoculating it into a plant, we just keep them from feeding deeply. We did not count the number of moribund GWSS, which would not be able to acquire or transmit the OLS bacterium

As an unanticipated benefit, Merit had an impact on infestation by the oleander aphid (*Aphis nerii*), an important pest of oleanders and other ornamentals (McAuslane 2001). In our bacterial inoculation experiment the oleander aphid infested 9.4% of untreated oleanders while none of the Merit-treated oleanders were infested.

Conclusions and Recommendations

We conclude that applications of Merit to oleanders would likely slow the spread of OLS among plants in two ways: (1) by killing potential GWSS vectors, and (2) by reducing the ability of the GWSS to feed and thereby limiting their ability to acquire the OLS bacterium from infected plants or inoculate it into non-infected plants. Our greenhouse and field studies consistently show that oleanders treated with Merit induce mortality in GWSS weather or not the plants are infected with *X. fastidiosa*. Our studies also show that Merit reduces feeding by the GWSS. This would limit the ability of non-infectious GWSS to acquire the pathogen from OLS-diseased oleanders, and for infectious GWSS to inoculate healthy oleanders with *X. fastidiosa*. Thus, Merit provides some level of protection in a "coming and going" fashion. It protects healthy oleanders from being inoculated, and infected oleanders from becoming sources from where the pathogen spreads. Because Merit has a long-lasting impact of over 3 months, few applications made during times when vector pressure is the greatest would optimize its utility. Unfortunately, our studies are not conclusive regarding the impact of Merit on transmission of *X. fastidiosa*. In spite of that, we recommend that Merit be used in oleanders that are under high risk of infection, or in circumstances where protecting them is paramount to maintaining important plantings of oleander.

We do not expect Merit to stop all spread of OLS, particularly in area of high disease pressure. Therefore, Caltrans should plan to replace oleanders in those areas with other ornamentals.

Implementation

The plant protection tactic discussed herein is implemented by applying Merit according to the manufacturers instructions to oleanders for which protection is desired. The choice of where and when to use it is critical. Because Merit is relatively expensive, and not necessary under all circumstances where oleanders are used, it should be used only in high risk areas were OLS is known to exist either in Caltrans right-of-ways or in nearby neighborhoods. OLS is especially common in coastal southern California (San Diego to Santa Barbara counties) and Riverside County, and parts of San Bernardino County. Although oleanders experience few other maladies that produce symptoms similar to OLS, commercial analytical laboratories (eg Agrianalysis, Davis CA) that examine plant material for *X. fastidiosa* can detect it with routine diagnostic assays.

At this point we do know what time of year infected oleanders are most likely to be so sources of *X. fastidiosa*, or when non-infected oleanders are most likely to be inoculated by infectious GWSS. However, we do know that GWSS is most active from approximately May through September (Fig 1) (Blua et al. 2001). Two half-rate applications may extend the timeframe of protection.

Figure1. Mean (+ standard error) of *Homalodisca coagulata* adults caught on yellow sticky cards from November 1996 through October 1999 on citrus ( ) and oleander ( ) in Irvine (**A**) and Riverside (**B**) California.

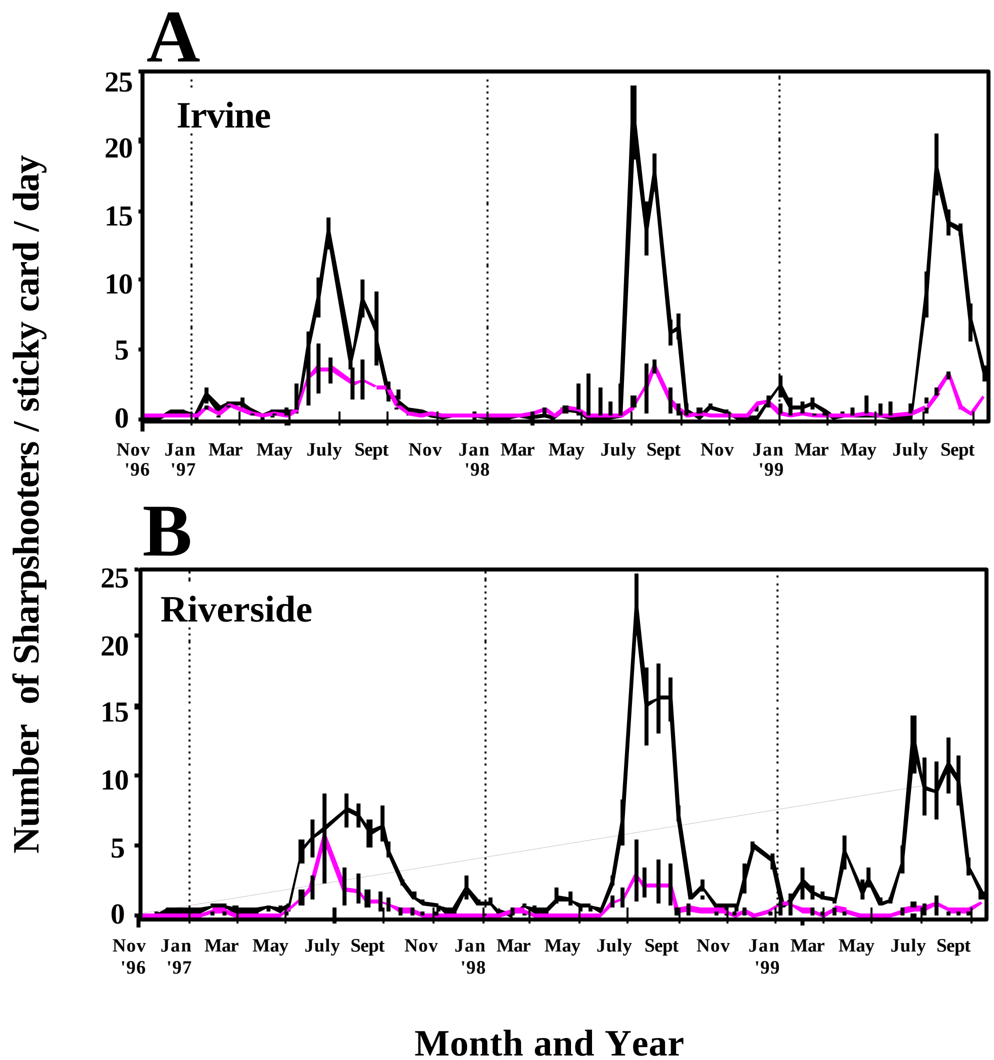


Figure2. Mean (+ standard error) of *Homalodisca lacerta* adults caught on yellow sticky cards from November 1996 through October 1999 on citrus (▬▬) and oleander (▬▬) in Palm Desert (**A**) and Riverside (**B**) California.

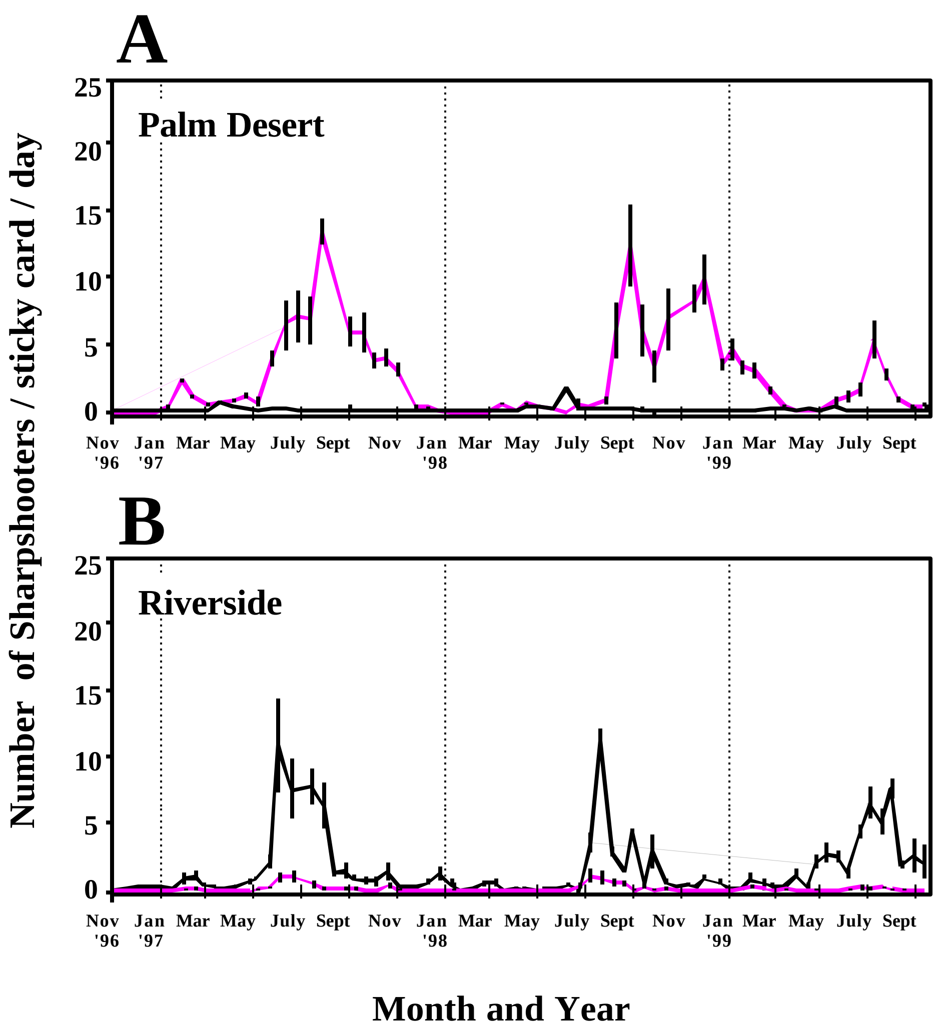


Figure 3. Percent mortality (mean $\pm$ 1 standard error) of *Homalodisca coagulata* 4 h after release on oleander treated with imidacloprid 0, 1, 2, and 3, wk earlier. Asterisk indicates statistically significant differences from control ($p < 0.05$).

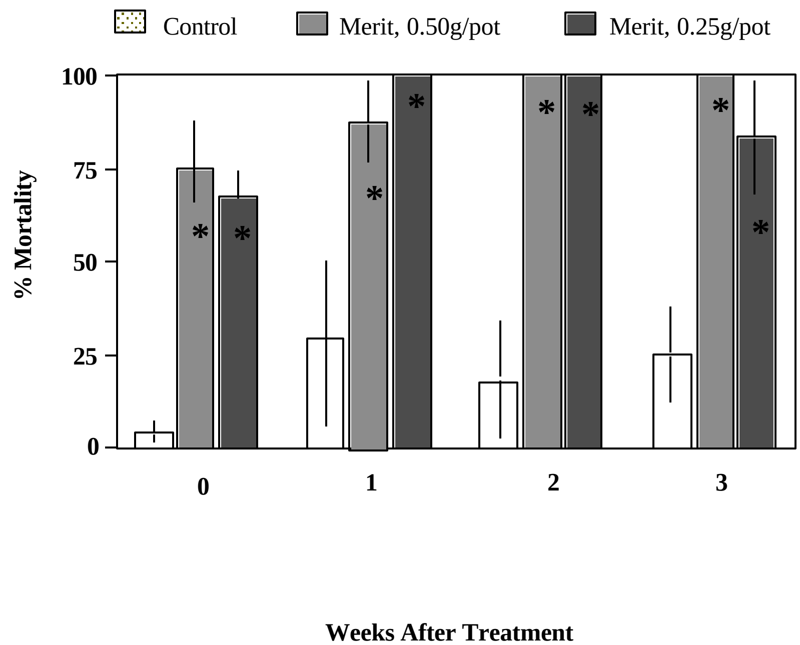


Figure 4. Minutes feeding (mean + 1 standard eror) (**A**), minutes to death (**B**), and percent OLS transmission (**C**), for *Homalodisca coagulata* individuals on treated oleanders and plants Treated one week earlier with imidacloprid. Letters above bars indicate statistically significant Differences ($p < 0.05$).

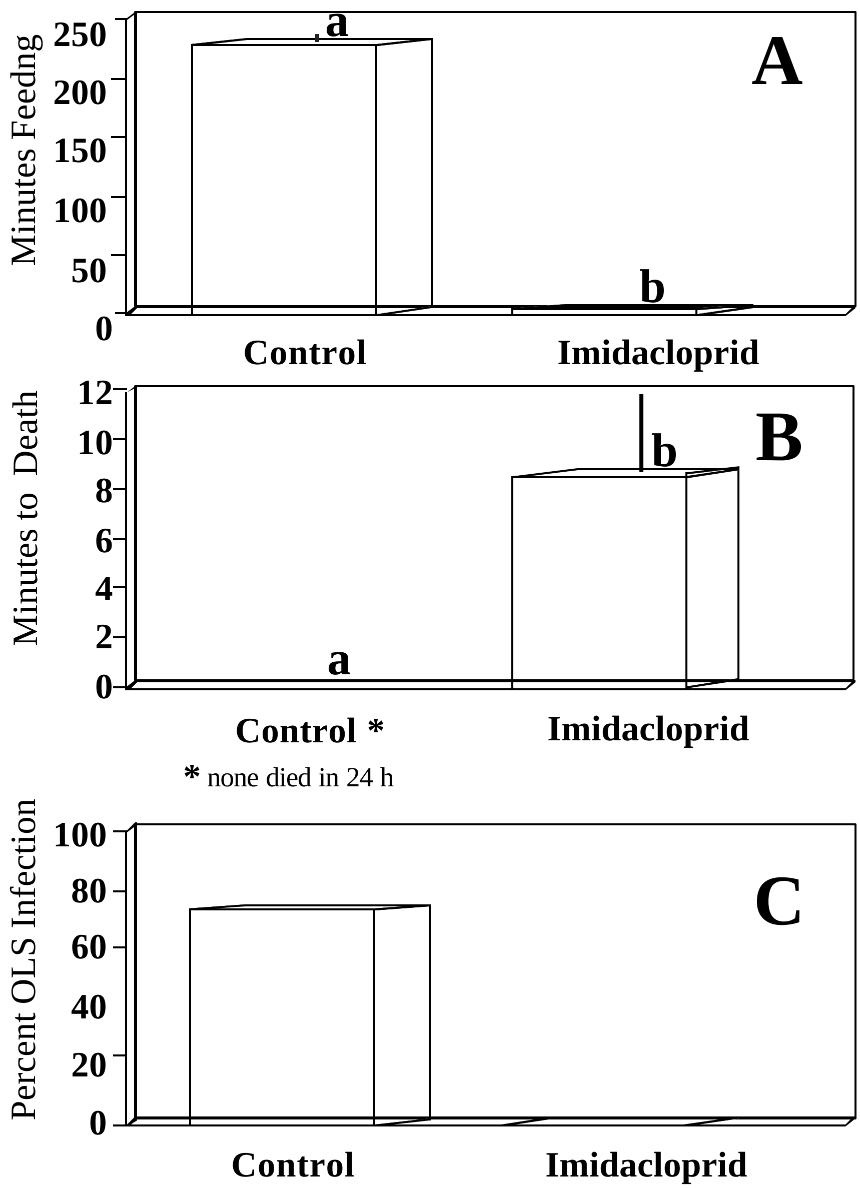
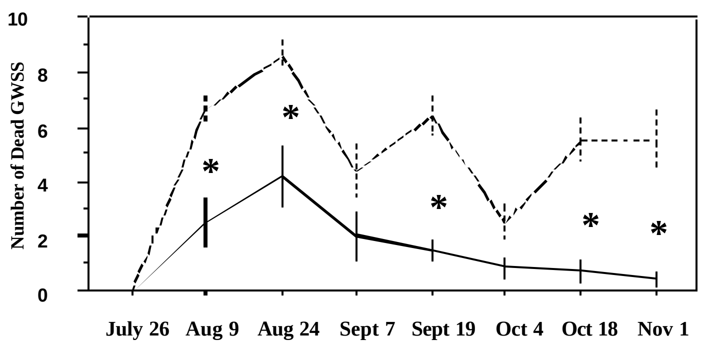
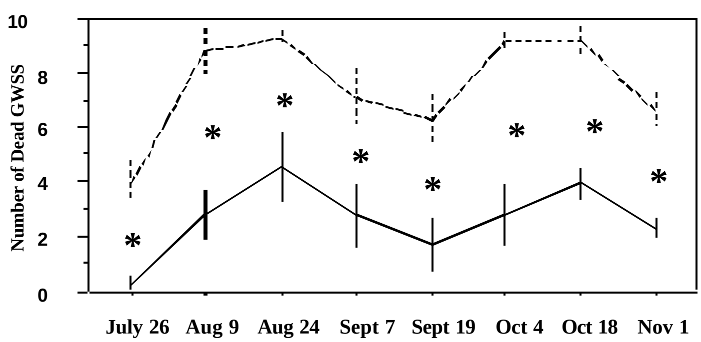


Figure 5. Mean number of dead *Homalodisca coagulata* on oleanders infected with *Xylella fastidiosa* and treated with Merit (dashed line), or left untreated (solid line). Treatment date was July 17, 2001. Graphs represent cucumulative mortality after 2, 4, and 6 hours of exposure to plants. Asterisks between points indicate stastically significant differences at $p < 0.05$.

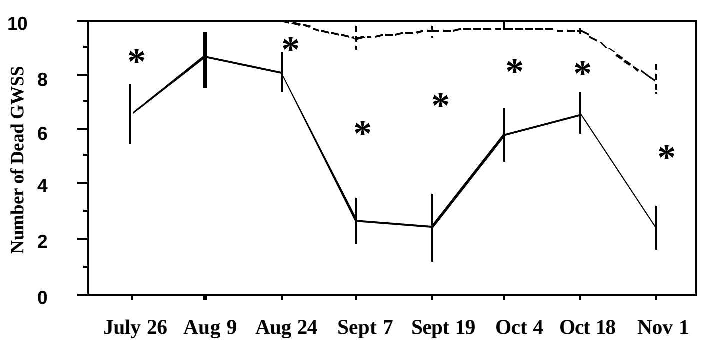
2 hr after exposure



4 hr after exposure



6 hr after exposure



Literature Cited

Adlerz, W. C., Bistline, F. W., Russo, L. W., & Hopkins, D. L. 1989. Rate of spread of citrus blight reduced when sharpshooter leafhoppers (Homoptera: Cicadellidae) are controlled. J. Econ. Entomol. 82, 1733-1737.

Anon. 1994. BAY NTN 33983. Technical and safety information sheets. Kansas City, MO, Mobay Ag. Chemicals Division.

Blua, M. J., R. A. Redak, D. J. W. Morgan, and H. S. Costa. Seasonal flight activity of two *Homalodisca* species (Homoptera: Cicadellidae) that spread *Xylella fastidiosa* in southern California. Accepted by Journal of Economic Entomology

Costa, H.S., Blua, M.J., Bethke, J.A., and Redak, R.A. 2000. Transmission *of Xylella fastidiosa* to oleander by the glassywinged sharpshooter, *Homalodisca coagulata*. Hortscience 35: 1265-1267.

Elbert, A., and Overbeck, H. 1990. Imidacloprid, a novel systemic nitromethylene analogue insecticide for crop protection. Proc. Brighton Crop Protection Conf. 2-1: 21-28.

Hendson, M, Purcell, A. H., Chen, D., Smart, C., Guilhabert, M., and Kirkpatrick, B. 2001. Genetic Diversity of Pierce's Disease Strains and Other Pathotypes of *Xylella fastidiosa*. *Applied and Environmental Microbiology* 67:895-903.

Henry, M., Purcell, S. A., Grebus, M., Blua, M. J., Hartin, J., Redak, R. A., Triapitsyn, S., Wilen, C., and Zilberman, D. 1997. Investigation of a new strain of *Xylella fastidiosa* & insect vectors as they affect California's agriculture and ornamentals industries. Technical report to the University of California Division of Agricultural and Natural Sciences. Grant # 113.

Hopkins, D. L. 1989. *Xylella fastidiosa*: xylem-limited bacterial pathogen of plants. Ann. Rev. Phytopathology. 27: 271-290.

McAuslane, H. 2001. Featured Creatures: Oleander Aphid, Aphis nerii Boyer de Fonscolombe. http://creatures.ifas.ufl.edu/orn/shrubs/oleander_aphid.htm.

Perring, T. M., Blua, M. J., Bethke, J., & Redak, R. A. 1997. Investigation of the effect of selected insecticides on sharpshooter vectors of oleander leaf scorch. Technical report submitted to CalTrans. No. FHWA/CA/OR-97-01.

Purcell, A. H., and Finlay, A. H. 1979. Evidence for noncirculative transmission of pierce's disease bacterium by sharpshooter leafhoppers. Phytopathology 69: 393-395

Sorensen, J. T., and Gill, R. J. 1996. A range expansion of *Homalodisca coagulata* (Say) (Hemiptera: Clypeorrhncha:Cicadellidae) to southern California. Pan-Pacific Entomol. 72: 160-161.

SAS Institute, 1999. StatView Reference. SAS publishing, Cary, NC.

Steel, G. D., and J. H. Torrie. 1980. Principles and procedures of statistics, a biometrical approach, second edition. McGraw-Hill, New York. 633pp.

Wesolowsk, G. O. 1976. Multiple regression and analysis of variance. John Wiley and sons, New York. 292pp.